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●*Original Contribution*

ULTRASONIC MYOCARDIAL INTEGRATED BACKSCATTER AND MYOCARDIAL WALL THICKNESS IN ANIMAL EXPERIMENTS

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Abstract—The purpose of this study was to distinguish between normal and ischemic myocardium using ultrasonic integrated backscatter (IB) measurements and to relate IB with myocardial wall thickness. IB was measured in 9 open-chested Yorkshire pigs (24–30 kg) before, after 30 minutes of partial occlusion of the proximal left anterior descending coronary artery (LADCA), and after 60 minutes of subsequent reperfusion. The ultrasound transducer (4 MHz) was sutured onto the epicardial surface perfused by the LADCA. IB measurements were made with a repetition rate of 50 times per heart rate simultaneously with a left ventricular pressure signal. Myocardial wall thickness was measured off-line. The measurements of integrated backscatter, left ventricular pressure and wall thickness were based on mean values of ten subsequent cardiac cycles. End-systolic IB measurements were 5.3 dB higher during occlusion as compared to the reference measurements (7.1 ± 3.2 dB versus 1.8 ± 2.6 dB; $p = 0.002$). No statistically significant differences were found in end-systolic IB measurements. End-systolic wall thickness was 5 mm smaller during occlusion as compared to the reference measurements (7.2 ± 1.4 mm versus 12.2 ± 1.2 mm; $p < 0.001$). Simple linear regression analysis showed a statistically significant inverse relationship between IB measurements and wall thickness in 21 out of the 23 sequences in which wall thickness could be measured. End-systolic IB measurements are favourable to distinguish acute ischemic myocardium from normal myocardium. There is a distinct inverse relationship between IB and myocardial wall thickness.

Key Words: **Ultrasound, Myocardium, Integrated backscatter, Wall thickness, End systole, End diastole.**

INTRODUCTION

Integrated backscatter is a relative measure of the ultrasonic energy backscattered by a small volume of tissue. Integrated backscatter provides a potentially useful index for quantitative ultrasonic characterization of myocardial tissue. In animal studies it has been shown that integrated backscatter is increased in acute ischemic myocardium as compared to normal myocardium (O'Donnell et al. 1979; Miller et al. 1985; Glueck et al. 1985).

Another promising application of integrated backscatter measurements is the assessment of the myocardial contractile function. Variation of integrated backscatter during the cardiac cycle has been reported in normal and ischemic myocardial tissue of dogs (Mottley et al. 1984; Wickline et al. 1985a; Wickline et al. 1985b) and in human subjects (Vered et al. 1989; Skorton 1989). In normal myocardium the diastolic value of integrated backscatter is higher as compared to the systolic value, whereas during acute ischemia this cyclic variation changes. Comparisons were made between the amplitude of cyclic variation of integrated backscatter and end-systolic percent myocardial wall thickening (Wickline et al. 1986).

The goal of this study was to explore the possibilities of integrated backscatter measurements to distinguish normal myocardium and acute ischemic myocardium in animal experiments. Given the cyclic variation of backscatter we wanted to know which instant of the cardiac cycle is optimal to make the distinction. Another question was whether the relationship between integrated backscatter and wall thickening is maintained throughout the cardiac cycle. For that purpose a computer-based measurement system was developed to record the broadband high frequency signals offering the possibility to analyze dynamic integrated backscatter and simultaneously acquired myocardial wall thicknesses.

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METHODS

General

Animal experiments were performed on Yorkshire pigs according to the guiding principles in the care and use of animals (Office of Science and Health Reports 1980). In order to provide adequate depth of anaesthesia, the animals (n = 9, 24–30 kg) were sedated with an intramuscular injection of 120 mg azaperone and subsequently, anaesthetized with 150 mg of metomidate which was administered via a dorsal ear vein. After intubation the animals were connected to a respirator for artificial ventilation with a mixture of 30% oxygen and 70% nitrous oxide. Anaesthesia was maintained with 160 $mg \cdot kg^{-1}$ alphachloralose (Merck, Darmstadt, FRG) followed by an infusion of 5 $mg \cdot kg^{-1}$ pentobarbitone sodium (Sonofi, Paris, France) via a catheter placed in the superior vena cava by way of a jugular vein. Respiratory rate and tidal volume were adjusted to maintain physiological arterial blood gas values (ABL-3, Radiometer, Copenhagen, Denmark). During the course of the present study, these values were 7.39 ± 0.01 (pH), 45 ± 3 mmHg (pCO_2), 154 ± 4 mmHg (pO_2) and 93 ± 2% (O_2 saturation), respectively.

Sodium bicarbonate (8.4%) and haemaccel (Behringwerke, Marburg, FRG) were infused to correct base deficit and blood loss. Left ventricular and central aortic pressure were monitored with microtipped catheters (Honeywell-Philips, Best, The Netherlands). After the heart was exposed using a midsternal split, a hydraulic occluder (RE Jones, Silver Spring, MD, USA) was placed around the proximal left anterior descending coronary artery (LADCA) distal and connected to a 1 mL syringe (Hamilton Bonaduz, Bonaduz, Switzerland) which was driven by a micrometer. During the study the chest of the pig was retracted (Sassen et al. 1988).

At the onset of occlusion, regional myocardial function was estimated from myocardial wall thickness recordings obtained with the aid of a 4 MHz ultrasonic transducer (Krautkramer-Branson, Lewistown, PA, USA) sutured into a part of the epicardial surface perfused by the LADCA which was connected to a commercially available echocardiographic ultrasound system (Organon Teknika, Oss, The Netherlands).

Experimental protocols

After cardiovascular parameters had been stable for at least 30 minutes following completion of the surgical procedures, baseline values of systemic haemodynamics, regional myocardial function and arterial blood gases were obtained. LADCA flow was then reduced gradually by slowly inflating the balloon until regional wall thickening had decreased to less than 20% of its baseline value.

If necessary, minor adjustments in the degree of flow reduction were performed during the first 5 minutes of ischemia, but, thereafter, no further manipulations were allowed. We have shown that after these 5 minutes perfusion and function of the ischemic and non-ischemic myocardium do not change further during the following 30 minutes (Sassen et al. 1988).

Data acquisition

Backscatter measurements were made at three episodes during the experiments: before the occlusion was applied, after 30 minutes of occlusion and after one hour of reperfusion in all pigs. The ultrasound transducer was connected to an in-house developed transmitter/wide band amplifier (input impedance 100 Ohm, fixed gain 29 dB, −6 dB cut-off frequencies 1.2 and 10 MHz). The amplified ultrasound signal was filtered by a 5th order 10 MHz low-pass Chebyshev filter and connected to one channel of a dual channel digital oscilloscope (LeCroy 9400).

Ultrasound signals from the time interval of interest (4.5–24.5 μs following the transmitter pulse) were digitized with 8-bit resolution at a sample frequency of 25 MHz providing for a Nyquist frequency of 12.5 MHz. At the same time a left ventricular pressure signal was digitized by the oscilloscope. The digital oscilloscope was interfaced with an IBM computer system (AT-3) which was described extensively elsewhere (Lancée et al. 1988).

Measurement sequences were generated by the computer system. The heart rate of the pig was entered into the computer and, starting at the onset of the pressure curve, 62 broadband transmitting pulses (approximately 1 μs) were generated with the repetition rate of 50 times the heart rate. After the last transmitting pulse the accumulated digitized ultrasound and pressure signals of at least one cardiac cycle were transferred to the computer system and displayed on a video monitor. Measurements were rejected by the operator when the time interval of the cardiac cycle did not match with a preset value of ±10%. This measurement procedure was repeated 10 times in order to obtain 10 useful cardiac cycles to be stored on disk. Usually this could be accomplished within 30–40 seconds. This procedure was repeated during the other episodes of the experiment.

Data processing

Data were processed off-line. Myocardial wall thickness was measured interactively using the com-

puter system. The recorded ultrasound signals of the complete cardiac cycles were displayed on the video monitor of the computer system in brightness mode using a 30 dB logarithmic signal compression. The endocardial wall of every cycle was traced continuously by an observer using a mouse (of the computer system). Only the endocardial walls which were clearly visualized were processed for further analysis.

In the next step the 10 consecutive cycles of every measurement were matched in time on the basis of the pressure curve. The individual pressure curves were displayed on the monitor and the beginning and end of the cardiac cycle were indicated by the observer. All time scales of the cardiac cycles were converted to a linear scale expressed as a percentage of the cardiac cycle ranging from begin systole (0%) to end diastole (100%) with 2% increments. Ultrasound spectra were calculated by a Fourier transformation using the array processor (Data Translation 7020) of the computer system. A split cosine bell window ($p = 0.1$; Bloomfield 1976) of 5–8 μs was chosen in order to exclude the endocardial wall signal from the spectra. Integrated backscatter was calculated by integrating the spectra from 2.5 to 7.5 MHz. The ultrasound data were not corrected for the frequency responses of the transducer and the wide band amplifier. All calculations were done using the same time window, resulting in the same, but arbitrary, 0 dB reference level for every backscatter measurement.

After completion of the data processing of the measurement sequences, the data of time dependent integrated ultrasound backscatter, wall thickness and left ventricular pressure at 2% increments of the cardiac cycle were available for each episode.

Mean values of the standard deviations of the integrated backscatter, wall thickness and left ventricular pressure were calculated for every 2% increment of the cardiac cycle in every pig. Plots were generated of the mean values and standard deviation versus time for visual inspection. End systole was defined as 30%–46% cardiac cycle and end diastole as being 80%–96%. End-systolic and end-diastolic measurements of integrated backscatter, wall thickness and left ventricular pressure were calculated by averaging the mean values of the parameters over the nine time intervals within the ranges of the respective definitions. Hence this averaged data were based on 90 measurements (10 cardiac cycles times 9 time intervals). This procedure was repeated for each episode.

Finally, end-systolic and end-diastolic mean values and standard deviations of every parameter were calculated to obtain the group values in the population of 9 pigs at basal state (the reference values), during occlusion and during reperfusion.

Statistical analysis

To explore the differences of the measurements obtained during three episodes of the experiment, the end-systolic and end-diastolic group values of the measurements obtained during occlusion and reperfusion were compared to the respective group reference measurements. An unpaired t test was applied and a p-value less than 0.01 was regarded as being significant.

The relationship between the integrated backscatter and myocardial wall thickness was explored in two ways. In order to investigate whether the inter-pig differences in integrated backscatter during the reference phase can be explained by differences in wall thickness, simple linear regression was performed between the reference measurements of integrated backscatter and the reference measurements of wall thickness. This was done for the end-systolic and end-diastolic mean values of the measurements separately.

Secondly, the dependency of the integrated backscatter on wall thickness was investigated in every separate measurement sequence. The 51 mean values of the time dependent integrated backscatter measurements were coorrelated with the mean values of the simultaneous wall thickness measurements. Simple linear regression analysis was applied using a significance level of $p < 0.01$.

RESULTS

Myocardial wall thickness could be measured in 8 pigs during the reference episode, in all pigs after 30 minutes occlusion, and in 6 pigs after 60 minutes reperfusion. In the other cases the endocardial wall was not clearly visualized.

A typical example of the time dependent measurements of integrated backscatter, myocardial wall thickness and left ventricular pressure during the reference episode and after 30 minutes of occlusion is shown in Fig. 1.

The obtained end-systolic and end-diastolic mean group values and the standard deviations during the three episodes are given in Table 1. The results of the integrated backscatter and wall thickness measurements obtained in end-systole and end-diastole during the three different episodes are graphically illustrated in Figs. 2 and 3.

End-systolic integrated backscatter during occlusion was significantly larger (5.3 dB) as compared to the end-systolic reference measurements. End-systolic wall thickness was significantly smaller (5 mm) during occlusion as compared to the end-systolic reference wall thickness. During reperfusion the left

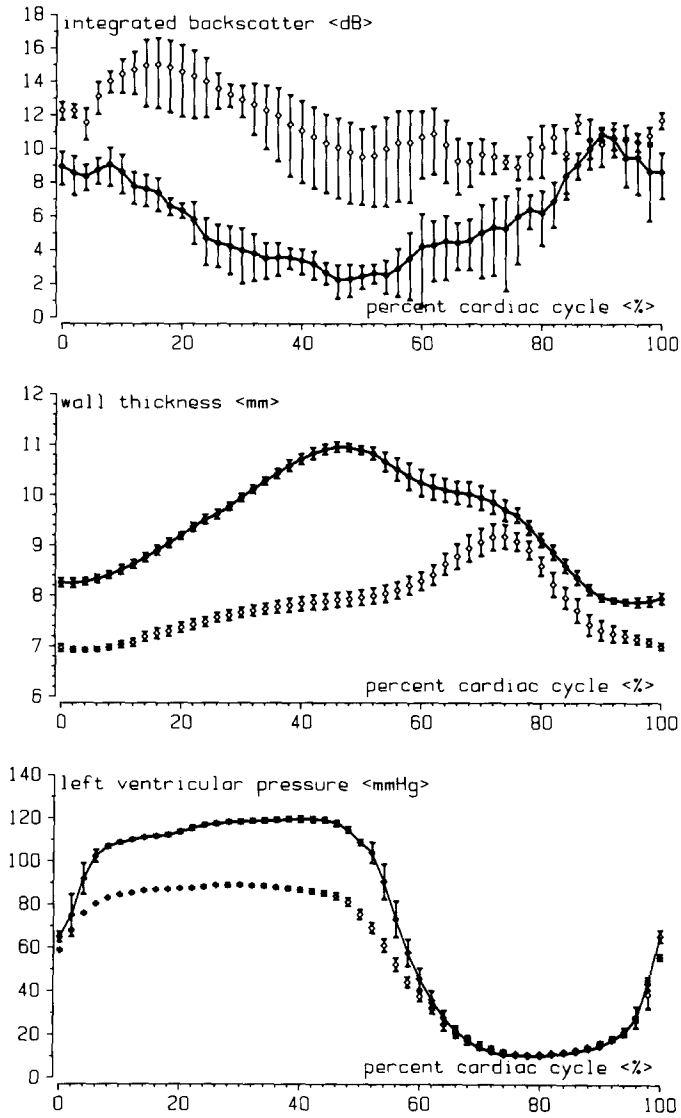

Fig. 1. The time dependent measurements of integrated backscatter (top), myocardial wall thickness (middle) and left ventricular pressure (bottom). Shown are the mean values (diamonds) and standard deviations (bars) of ten cardiac cycles in pig no. 8 during the reference episode (connected with a solid line) and after 30 minutes occlusion.

ventricular pressure was significantly lower (35 mmHg) as compared to the pressure during the reference episode.

No significant differences were found comparing the end-diastolic integrated backscatter measurements during occlusion and reperfusion with the respective reference measurements. A small, but significant, difference (1.5 mm) was found comparing end-diastolic wall thickness measurement during occlusion with the reference measurement.

No significant relationship was found between the mean values of integrated backscatter and wall thickness measured during the reference episodes, both in end-systole and end-diastole. Therefore, the inter-pig differences in integrated backscatter could not be explained by differences in wall thickness.

The results of the simple linear regression analysis of the integrated backscatter and wall thickness of the 23 available sequences in which wall thickness could be measured showed significant inverse relationships between integrated backscatter and wall thickness in all but two cases. In one sequence measured during occlusion there was a nonsignificant inverse relationship and in one reference sequence the integrated backscatter increased significantly with increasing wall thickness. Typical examples of the scatterplots are shown in Fig. 4 (reference) and Fig. 5 (after 30 minutes occlusion).

Table 1. Group mean values and standard deviations (between brackets) of integrated backscatter (IB), wall thickness and left ventricular (LV) pressure, in end systole (top) and end diastole (bottom) during the three different episodes of the experiments.

	Reference	Occlusion	Reperfusion
End systole			
IB (dB)	1.83 (2.62)	7.10 (3.15)*	3.51 (2.50)
Wall thickness (mm)	12.2 (1.18)	7.22 (1.43)*	10.8 (2.29)
LV pressure (mmHg)	95.0 (16.3)	73.3 (19.9)	59.8 (21.5)*
End diastole			
IB (dB)	4.67 (3.57)	5.63 (4.25)	3.74 (4.05)
Wall thickness (mm)	9.48 (1.24)	7.84 (1.05)*	10.24 (2.53)
LV pressure (mmHg)	13.8 (6.12)	15.13 (4.83)	13.7 (6.14)

An asterisk indicates a significant difference ($p < 0.01$, unpaired t test) as compared to the respective reference measurements.

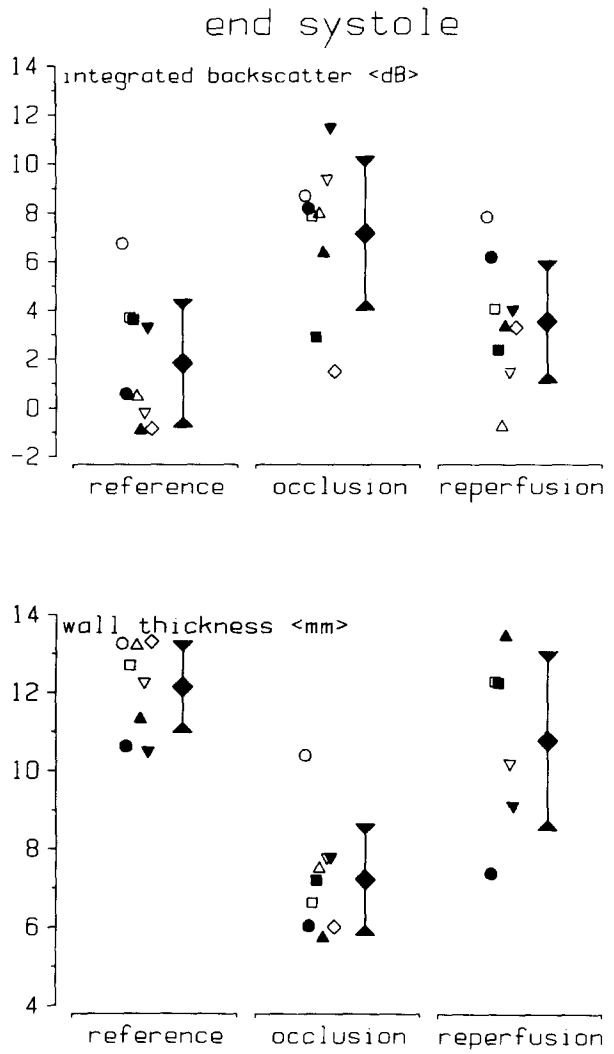

Fig. 2. End-systolic mean values of integrated backscatter (top) and wall thickness (bottom) obtained during the different episodes of the experiment. Shown are the mean group values (solid diamonds) and standard deviations (bars). The different symbols indicate the measurements obtained in the different pigs.

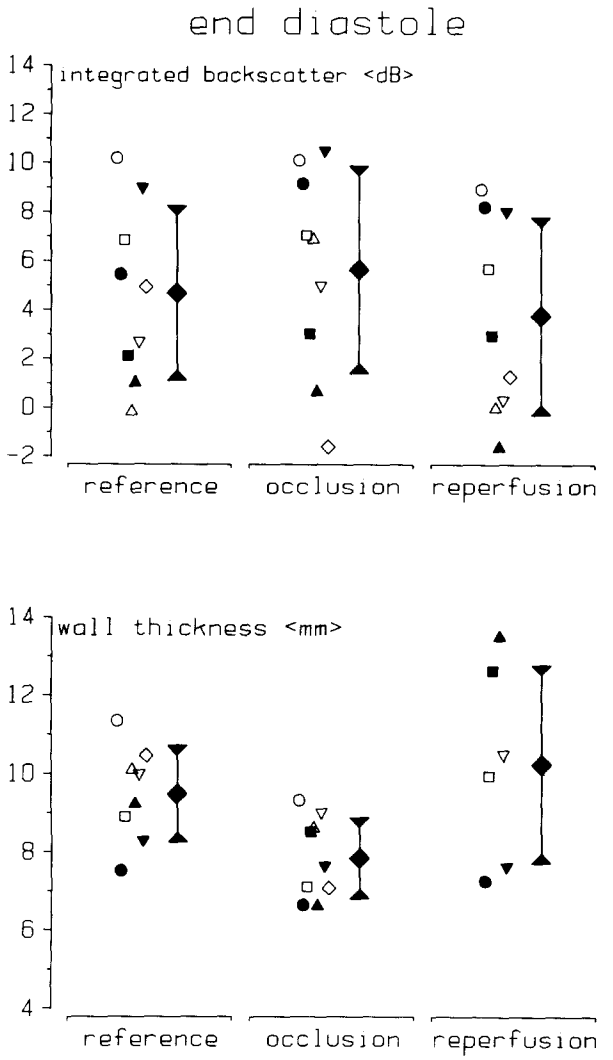

Fig. 3. End-diastolic mean values of integrated backscatter (top) and wall thickness (bottom) obtained during the different episodes of the experiment. Shown are the mean group values (solid diamonds) and standard deviations (bars). The different symbols indicate the measurements obtained in the different pigs.

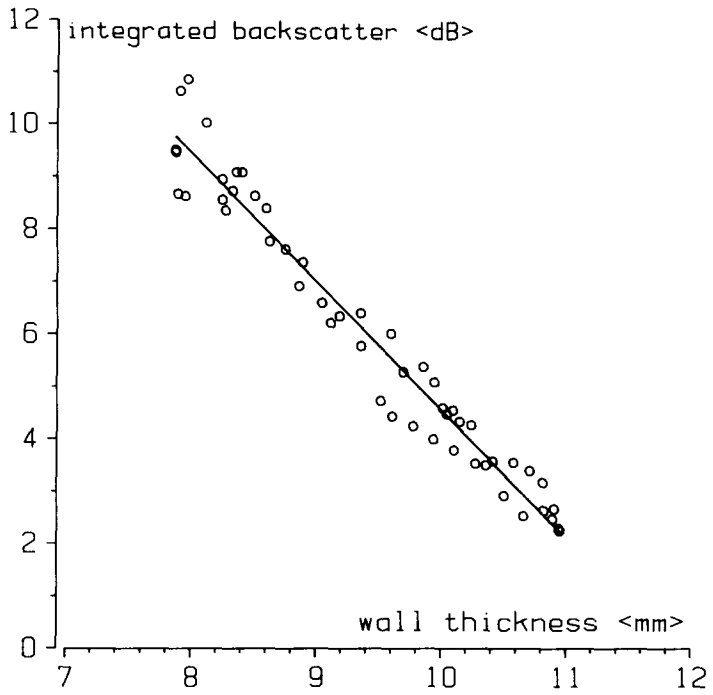

Fig. 4. Scatterplot of the integrated backscatter measurements versus myocardial wall thickness ($R = 0.98$, $p < 0.01$). The open circles depict the paired mean values of ten cardiac cycles during the reference episode of pig no. 8.

DISCUSSION

An important clinical application of myocardial ultrasonic integrated backscatter is the distinction between normal myocardium and ischemic myocardium. Our experiments show that end-diastolic integrated backscatter measurements could not distinguish between normal myocardium and acute ischemic myocardium.

End-systolic integrated backscatter measurements showed more favourable perspective for this application of cardiac tissue characterization. In our experiments the end-systolic integrated backscatter was 5.3 dB higher during occlusion as compared to the end-systolic reference measurements. However, due to the overlap of the measurements it would be difficult in some cases to distinguish normal myocardium from occluded myocardium.

An important observation is the large variation (range ± 5 dB) of the integrated backscatter of the different myocardia of the various pigs, which could not be explained, even in part, by the differences in wall thickness. At this stage this variation can only be labelled as inter-pig variation, which must be subject for further investigation. A fairly large inter-subject variation would be a serious drawback for the application of tissue characterization.

The other subject of the study was to explore the relationship between integrated backscatter and myocardial wall thickness throughout the cardiac cycle. Since 21 out of 23 measurement sequences shows a statistically significant inverse relationship, there is evidence that the integrated backscatter increases with decreasing wall thickness. These observations were made during the separate episodes of the study and cannot be applied to explain the increase in end-systolic integrated backscatter by the decrease of end-systolic myocardial wall thickness after occlusion of a coronary artery. It should be realized that after the occlusion of a coronary artery two factors might influence the integrated backscatter change simultaneously: the myocardial wall thickness decreases and the myocardial tissue enters the state of acute ischemia. During ischemia the end-diastolic wall thickness was only 1.6 mm smaller as compared to the reference thickness, whereas end-systolic wall thickness was 5 mm smaller as compared to the baseline measurements.

This larger end-systolic difference in wall thickness might explain part of the end-systolic increase in backscatter measurements during ischemia, given the observed relationship between backscatter and wall thickness. Another part may be explained by the state of acute ischemia of the myocardium. Since the variables "ischemia" and "wall thickness" are not dependent one cannot assess their relative contribution to

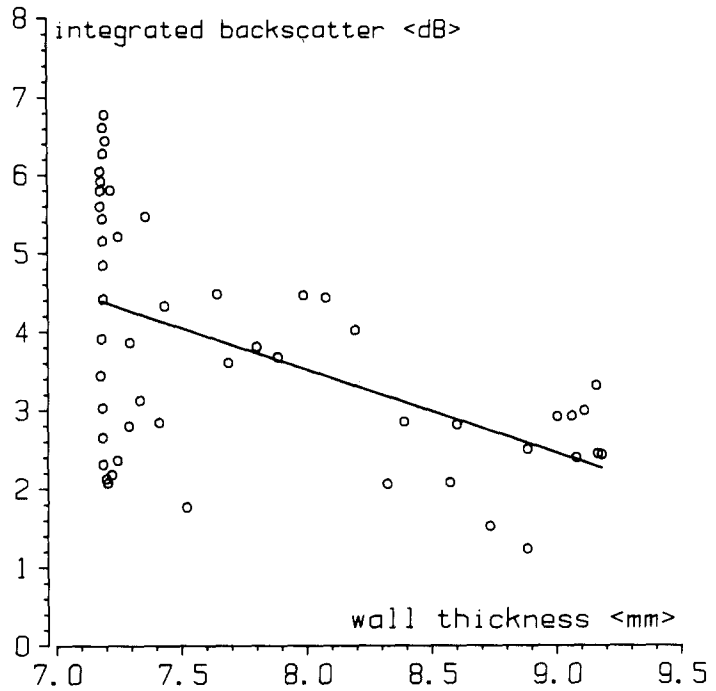

Fig. 5. Scatterplot of the integrated backscatter measurements versus myocardial wall thickness ($R = 0.52$, $p < 0.01$). The open circles depict the paired mean values of ten cardiac cycles after 30 minutes occlusion of pig. no. 1.

the increase in end-diastolic integrated backscatter very accurately. To avoid this problem, one has to compare normal and ischemic myocardial tissue having the same ranges of wall thickness. Theoretically, this can be done by comparing reference measurements of backscatter and wall thickness with the reperfusion measurements in those pigs with persisting ischemia during reperfusion. Given the small number of available measurements (in our study only 5 pigs had measurable wall thicknesses during reperfusion and did not recover from ischemia) and the large inter-subject variation of the backscatter measurements the results in this study would be inconclusive.

An inverse relationship between integrated backscatter and myocardial wall thickness confirms the reported cyclic variation of myocardial integrated backscatter in dogs (Mottley et al. 1984; Wickline et al. 1985a; Wickline et al. 1985b), although these measurements were obtained from the focal zone of the transducer at 5 cm distance. Our measurements were obtained in the near field of the transducer. In this situation phase cancellations at the aperture of the transducer may have influenced our measurements. It has been shown (Johnston and Miller 1986) that phase cancellations will cause fluctuations in the backscatter measurements and a lower absolute level of integrated backscatter as compared to phase insensitive measurements. Even though phase cancellations cannot be neglected, they do not seem to affect the cyclic variation of integrated backscatter measurements greatly. An essential point of this investigation is that all analyses were done with the same time-window at the same distance from the transducer, so that measurements could be compared and differences in measurements could not be explained by differences in phase cancellations.

CONCLUSION

On the average, end-systolic integrated backscatter in acute ischemic pig myocardium measured to be 5 dB higher as compared to end-systolic backscatter measurements in normal myocardium. Therefore, end-systolic integrated backscatter measurements may be able to distinguish normal myocardium from acute ischemic myocardium contrary to end-diastolic backscatter measurements.

There is a significant inverse relationship between the myocardial integrated backscatter and the myocardial wall thickness throughout the cardiac cycles of the various episodes (reference, occlusion and reperfusion) and shows integrated backscatter measurements to be an indicator of myocardial contractile performance. Whether this relationship explains in full the increase in end-systolic integrated backscatter after an occlusion is still under investigation.

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